PATENT
3920-0110P

IN THE U.S. PATENT AND TRADEMARK OFFICE

Applicant: John CARTER

Appl. No.: 10/089,846 Group: 1616

Filed: June 6, 2002 Examiner: Choi

For: PHARMACEUTICAL COMPOSITIONS AND THEIR USE IN THE TREATMENT OF NEOPLASTIC DISEASE

DECLARATION UNDER 37 CFR 1.132
(#1)

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

I, Roger Anthony Oakes, do hereby declare and state as follows:

1. I am Roger Anthony Oakes and am employed by Ivy Medical Chemicals Ltd., 54 Sun Street, Waltham Abbey, Essex, EN9 1EJ, UK, as Scientific Director.
2. I have read and am familiar with the specification and claims of the above-identified patent application.
3. In order to confirm the advantages of the invention described and claimed in the above application, the following studies were conducted under my supervision and control.

4. The study was conducted to assess the anti-tumor activity of the composition CV247 and combinations of the constituents thereof against LL2/LLc1 tumors in C57BL mice, Lewis Lung Carcinoma (LLc). The effect of the active agents was compared to untreated control animals bearing such tumors.

5. The CV247 composition comprised the following components:

Sodium salicylate	3.5 mg/mL
Ascorbic acid	4.0 mg/mL
Copper gluconate	0.2 mg/mL
Manganese gluconate	0.2 mg/mL

6. Gemcitabine, a highly active anti-tumor agent, was used as a control, as Gemcitabine has been shown previously to be effective against LLc grafts, and was thus deemed to be a positive control in the studies.

7. LL2/LLc1 cells were injected subcutaneously into female C57BL6/mice. A total of 110 mice were allocated to 10 dose groups. A first group assessed a dosing regimen of CV247 administered at 10mL/kg daily from the day of tumor inoculation, for a period of 21 days. All other regimens commenced 7 days after inoculation. In second, third and fourth groups, CV247 was dosed daily at 3mL/kg, 10mL/kg and 20mL/kg, respectively, for 14 days. Groups five to eight looked at the components of CV247, sodium salicylate (35mg/kg), sodium salicylate + ascorbic acid (35mg/kg + 40mg/kg), sodium salicylate + ascorbic acid + copper gluconate (35mg/kg + 40mg/kg + 2mg/kg), or sodium salicylate + ascorbic acid + manganese gluconate (35mg/kg + 40mg/kg + 2mg/kg), being dosed once daily for 14 days. In group 9 Gemcitabine was administered every

3rd day on 5 occasions commencing one week after tumor inoculation. Group 10 was left untreated. CV247 and its respective components were administered by oral gavage. Gemcitabine was administered via intraperitoneal injection.

8. Twenty-one days after tumor inoculation all animals were sacrificed. Tumors were excised, weighed and evaluated macroscopically. In addition, the tumors from the CV247 (10mL/kg), SS+AA, SS+AA+MG, Gemcitabine, and untreated control groups were subjected to histological scoring.
9. Based on the above evaluation, it was clear that the respective tumor weights demonstrated a significant difference between those treated with the CV247 composition and the untreated control. The respective tumor weights are depicted in attached Figure 1 (Mean tumor weights).
10. Macroscopic examination of tumors on excision revealed a difference in tumor structure between the respective treatment groups. Tumors treated with CV247 and SS+AA+MG appeared to be fluid-filled and spongy in comparison to the untreated controls and those treated with the other components. Gemcitabine-treated tumors were slightly smaller and did not appear to contain much fluid.
11. With regard to the histological scoring, tumor tissue (formalin fixed) from each animal was bisected, processed, embedded and sectioned for haematoxylin and eosin staining. Each slide was examined for the extent of intra-tumoral necrosis, and a score assigned as follows:

0	No necrotic foci
1	<25% of the area of the tumor is neocrotic
2	25-50% of the area of the tumor is neocrotic

3 >50% of the area of the tumor is neocrotic

The resulting mean group scores for intra-tumoral necrosis were:

Untreated	1.7
Gem	1.11
SS+AA+MG	1.25
SS+AA	1.37
CV247	1.2

12. The study accordingly demonstrates the effectiveness of the composition CV247 as an anti-tumor agent. The decrease in tumor weight indicates the existence of a mechanism for tumor reduction that is not necessarily related to tumor volume.

13. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under 18 U.S.C. 1001 and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Dated: 18th June 2008 Signed: _____

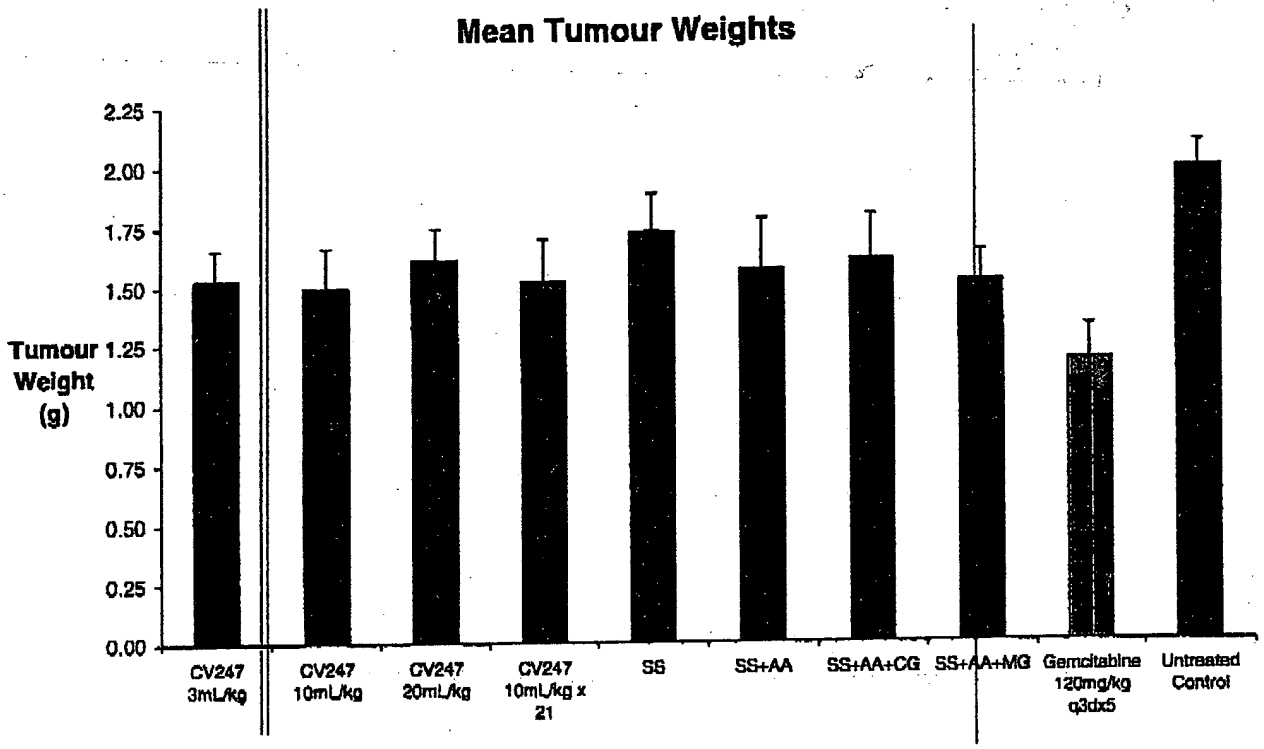
Mean Tumour Weights
Tumour Weight (g)
2.25
2.00
1.75
1.50
1.25
1.00
0.75
0.50
0.25
0.00
CV247 3mL/kg
CV247 10mL/kg
CV247 20mL/kg
CV247 10mL/kg x 21
SS
SS+AA
SS+AA+CG
SS+AA+MG
Gemcitabine 120mg/kg q3dx5
Untreated Control

FIGURE 1